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l1 same (genetic near0 mapping)	5

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L15: Entry 1 of 5 File: USPT Sep 12, 2000

DOCUMENT-IDENTIFIER: US 6117634 A
TITLE: Nucleic acid sequencing and mapping

DEPR:
In one embodiment, SR synthesis initiates at a unique site using an excess of processive polymerase, which incorporates DATP, dGTP, dCTP, dUTP (or any other labile base) into the DNA (FIG. 8). After a controlled period of incorporation of the labile base, conditions are changed to incorporate only the stable bases DATP, dGTP, dCTP, and labeled dTTP. The dTTP can be radioactively labeled, fluorescently labeled, chemically labeled with biotin, etc. The uracil bases can be removed using dU glycosylase (Boehrenger Meinheim), and the sites efficiently converted to nicks by heating the DNA. After cleavage of the dUTP-substituted DNA, the labeled DNA from the different SR reaction times can be hybridized to a sequence of interest (e.g., telomeric sequences, dinucleotide repeats, alu sequences, cloned or PCR-amplified sequences, expressed sequences from a cDNA library, etc.). Either the strand replacement DNA or the sequences of interest can be labeled. In the example shown in FIG. 8, positive hybridization would be detected for the samples from SR reactions carried out for about 15 min, 20 min, and 30 min. If the measured rate of SR elongation was 250 nucleotides per minute, those features would be mapped as being 3.75 kb, 5.0 kb, and 7.5 kb from the initiation site. By hybridizing to restriction fragments transferred from an agarose gel, the order of the restriction fragments could be easily mapped. This information could be very useful in large-scale sequencing projects to order the restriction fragments in cosmids and YACs. As the time increases the polymerases will lose synchrony and the width of the band of stable DNA will increase, reducing resolution. To overcome this problem agents can be introduced to reversibly halt the polymerase molecules at specific sequences. When the arrest is reversed all of the polymerases will regain their initial synchrony. Triplex-forming oligonucleotides can bind to recognition sequences along DNA and can arrest the progress of Klenow fragment [Hacia et al., "Inhibition of Klenow fragment DNA polymerase on double-helical templates by oligonucleotide-directed triple-helix formation," Biochemistry 33:6192 (1994)]. The arrest by oligonucleotides should be reversed by mild heating or changes in pH.

L11: Entry 9 of 196 File: USPT Apr 24, 2001

DOCUMENT-IDENTIFIER: US 6222103 B1
TITLE: Inbred maize line PH45A

DEPR:
In addition to phenotypic observations, the genotype of a plant can also be examined. There are many laboratory-based techniques available for the analysis, comparison and characterization of plant genotype; among these are Isozyme Electrophoresis, Restriction Fragment Length Polymorphisms (RFLPs), Randomly Amplified Polymorphic DNAs (RAPDs), Arbitrarily Primed Polymerase Chain Reaction (AP-PCR), DNA Amplification Fingerprinting (DAF), Sequence Characterized Amplified Regions (SCARs), Amplified Fragment Length Polymorphisms (AFLPs), and Simple Sequence Repeats (SSRs) which are also referred to as Microsatellites.

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End of Result Set

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L15: Entry 5 of 5 File: USPT Jan 9, 1996

DOCUMENT-IDENTIFIER: US 5482836 A
TITLE: DNA purification by triplex-affinity capture and affinity capture electrophoresis

DEPR:
These plasmids were then tested to determine whether or not they contained (dT-dC).sub.n .multidot.(dG-dA).sub.n sequences using a PCR-based method originally developed by Kunkel and coworkers (Feener, C. A., Boyce, F. M. & Kunkel, L. M. (1991) Am. J. Hum. Genet. 48:621-627). In this assay, a (T-C).sub.n, oligonucleotide and one of the sequencing primers were combined and used as PCR primers. If the tested insert contains a (dT-dC).sub.n .multidot.(dG-dA).sub.n sequence(s), amplified products should be obtained by, at least, one of the primer combinations depending on the relative orientation of the dinucleotide repeat. In practice, 17 clones of the 18 tested gave distinct PCR products, part of which are shown in FIG. 3. The results demonstrated that the triplex-mediated procedure is quite effective and useful for selection of (dT-dC).sub.n .multidot.(dG-dA).sub.n dinucleotide repeat clones and, thus, for the subsequent development of highly informative DNA markers for genetic linkage mapping. (FIG. 3 shows the results of the PCR-based assay for the (dT-dc).sub.n .multidot.(dG-dA).sub.n dinucleotide repeats on six of the eighteen randomly chosen clones (nos. 1-6) purified by TAC from a chromosome 21-specific library. PCR was performed with forward and reverse primers (each left lane), forward and BamTC (SEQ ID NO:2) primers (each middle lane), and reverse and BamTC (SEQ ID NO:2) primers (each right lane). Lanes M contain phage .lambda. DNA digested with HindIII (outside) and BstEII (inside)).